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Lung Model Casting Techniques for Interspecies Morphometric Comparisons

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LUNG MODEL CASTING TECHNIQUES FOR INTERSPECIES MORPHOMETRIC COMPARISONS

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ABSTRACT

Techniques have been developed for casting both solid and hollow lung models from lung specimens. These techniques have been used to make casts of rat, rabbit, baboon, and human lungs and may be used for other species. An air line at a positive pressure of 25 cm of water is connected to the trachea to air-dry lungs that have been surgically removed from the species. To make a solid model, silicone rubber is injected into the dry lung. Solid models also can be made by injecting silicone rubber directly *in situ* using saline displacement techniques. Nasopharynx casts are made by direct

injection of silicone rubber through the trachea. Using the lost wax technique, a hollow cast of the airways and the nasopharynx can be made. Anatomical differences in lungs and nasal passages have been observed among species. Current research investigates casting of the respiratory tract starting at the nares and extending through the larynx to the tracheobronchial tree.

KEY WORDS: Solid and hollow lung casts, breathing lung models, nasopharynx casts

INTRODUCTION

When particles are inhaled, their aerodynamic properties, combined with various anatomical and breathing characteristics, determine their fractional deposition in various regions of the respiratory tract. Mathematical models that predict pulmonary dose patterns of inhaled particles are useful in assessing health effects that are observed in animal experiments and in predicting effective pollutant concentrations between animals and man. Using available deposition data, the Task Group on Lung Dynamics of the International Radiological Protection Commission (1) developed a mathematical model for the deposition of insoluble nonhygroscopic particles in the human respiratory tract. More recently, the regional deposition in normal adult humans of monodisperse, insoluble, stable aerosols of different size has been studied extensively by Stahlhofen et al. (2), Heyder et al. (3), Chan and Lippmann (4), Davies (5), Pattle (6), Schlesinger and Lippmann (7), and Yu et al. (8). Raabe et al. (9) developed analogous deposition curves for animals. Recently, Martonen (10) developed an analytical model of hygroscopic particle behavior in human airways.

Upper respiratory tract morphometric data are limited (11,12). However, lower respiratory tract morphometric data have been provided by preparing and measuring replica casts. Solid casts were used by Weibel (13), Horsfield and Cumming (14), and Raabe et al. (15) to study conducting airway geometries, while Schreider and Raabe (16) examined the structure of the pulmonary acinus. Phalen et al. (17) developed a method for casting lungs in situ. Kliment (11) used latex to produce flexible solid lung casts for morphometric measurements. Preparation of a hollow cast was described by Timbrell et al. (18) and modified by Schlesinger and Lippman (19) for particulate deposition studies. The diameters, lengths, and branching angles of the conducting airways were shown to be different for various species (15,20).

Most anatomical respiratory tract models used for deposition studies have illustrated the tracheobronchial region (19,21) without the larynx or nasal passages in the system. Proetz (22) was one of the first investigators to consider the laryngeal jet in his analysis, and he observed that areas of turbulence caused by the jet impaction corresponded with higher deposition of particles. Chan et al. (23,24) reported the effects of the larynx on deposition patterns. Casting of the larynx has been reported by Schlesinger et al. (25). Martonen and Lowe (26) used rubber casts of human larynxes obtained at autopsy to study deposition of monodisperse fluorescein aerosols. All these studies show a marked difference in deposition patterns when the upper respiratory region was considered.

An exact anatomical model of the nose, larynx, and tracheobronchial tree is needed to describe the fluid flow characteristics of the lung, understand differences among species, and predict deposition of particles. In this report, casting of the respiratory tract starting at the nares and extending

through the larynx to the tracheobronchial tree is discussed. Silicone rubber casts were prepared from the total respiratory tract and measurements were made in various species. Possible relations between species are discussed. A method to obtain hollow casts is described.

MATERIALS AND METHODS

Flexible silicone rubber casts of the total respiratory tract were made by injecting silicone rubber (Dow Corning 734 RTV®, Dow Corning, Midland, MI) into the system according to the method described by Patra et al. (27). The nasal passages and tracheobronchial tree were cast separately. The different species used are listed in Table 1. To make solid casts of the nasal-pharyngeal airway, the trachea of the specimen was opened by tracheotomy. The mucus was washed out of the respiratory system by forcing normal saline through the airway. Trichloroethylene was added to self-leveling adhesive silicone (Dow Corning 734 RTV®) in a 1 to 10 ratio (by volume) for easy dispensing. This mixture was introduced through the trachea, up through the nasal cavity, and out through the external nares by means of a graduated dispensing cylinder at a pressure of 70 kg/m^2. The silicone rubber was allowed to cure *in situ* for 120 h. When the silicone casts were cured, they were surgically removed. The excess tissue surrounding the cast was digested by boiling the cast in a sodium hydroxide (NaOH) solution. After digestion, the solid model was used as a core to make a mold of the nose, mouth, nasopharynx, and larynx.

The lung casts used in this study were made by surgically removing the lung from the specimen. Dry air at a positive pressure of 25 cm of water was passed through the lung for 72 h to air-fix the lungs. Silicone rubber was

injected through the trachea by using a dispensing cylinder according to the pressure program reported by Patra et al. (27). When the silicone-filled lungs were cured, they were digested in boiling 1 N NaOH solution. After the tissue was digested, the solid models were washed and dried. To allow accurate measurements, the casts were trimmed to the terminal bronchiolar level. A Spencer dissecting scope (American Optical Company, Buffalo, NY) with a low magnification lens (10X, Leitz, Wetzlar) was used to measure the dimensions of the airways.

Hollow casts were prepared by using the solid lung models. The tracheobronchial casts were attached to the nasopharyngeal casts and placed in a mold box. Mold-making silicone rubber (Dow Corning SILASTIC® E-RTV) was used to make a sandwich mold. When all of the solid model was covered with the layers of silicone rubber and curing was complete, the mold was carefully disassembled to permit removal of the solid silicone core. Then the mold was reassembled, and hot liquid red Freeman wax was poured into the mold cavity and allowed to solidify. The mold was disassembled again, and the wax model was removed and coated with silicone rubber. When the coatings were cured, the wax was boiled off from the hollow model by the lost wax process.

RESULTS AND DISCUSSION

Anatomical differences in lungs were observed among species. Figure 1 shows models of some of the species investigated in this study, and Table 2 contains morphometric data for some species. Visual examination of a rat lung cast reveals that the lung is monopodial. The left lung has only one lobe, while the right lung has four lobes. Rat lung lobes have short lateral branchings that are attached with long tapering major bronchi at relatively

large angles. From a superficial anterior view, the human and baboon lungs appear quite similar. However, closer inspection shows that the human lung structure is very different from that of the baboon lung. The baboon lung has three lobes on the left side instead of the two in the human lung. The right side of the baboon lung has one more lobe than that side of the human lung. In contrast, the dog lung has three symmetric lobes on each side with a long trachea. A golden hamster lung has six lobes: two in the left lung and four in the right lung. This lung configuration is different from that of the rat or mouse, but is similar to that of the rabbit.

The baboon lung is an irregular dichotomus type, as is the human lung, while rat, hamster, and mouse lungs are monopodial. The dog lung and rabbit lung are combinations of both types; they are dichotomous in the upper airways and monopodial in the lower airways. The length of the trachea is dependent on the species. Human and baboon tracheas are relatively short when compared to rat or dog tracheas.

The various nasal-pharyngeal airway casts shown in Figure 2 are consistent with those made by other investigators (28,29,30). Casts from the same species were nearly identical with the exception of their size. However, a number of differences were observed among species. The length of the nasopharynx relative to the total nasal length was longest in rodents and shortest in humans. This measurement in the rabbit and dog was between that for the rodent and that for the human. The maxilloturbinate region in the human and baboon was fairly simple, but it was extremely complex in the dog. The rabbit ethmoturbinate region had layers of membranous chambers unlike those in other species. The guinea pig had an ethmoturbinate region that was similar, but not as complex, as that of the rabbit. The nasopharynx of humans

and monkeys has a sharp bend because of the upright posture of these species. The larynx and laryngopharynx were relatively longer in the rat and mouse as compared to these areas in the monkey and human.

In conclusion, silicone rubber casts can be used to evaluate and understand anatomical differences among species. The difference in number of lobes in each species, the branching patterns of different airways, and their morphometric dimensions can be studied with casts. Extrapolation of different respiratory tract characteristics from one species to another must be carefully examined because simple scaling may not be sufficient to produce accurate conclusions.

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DISCLAIMER

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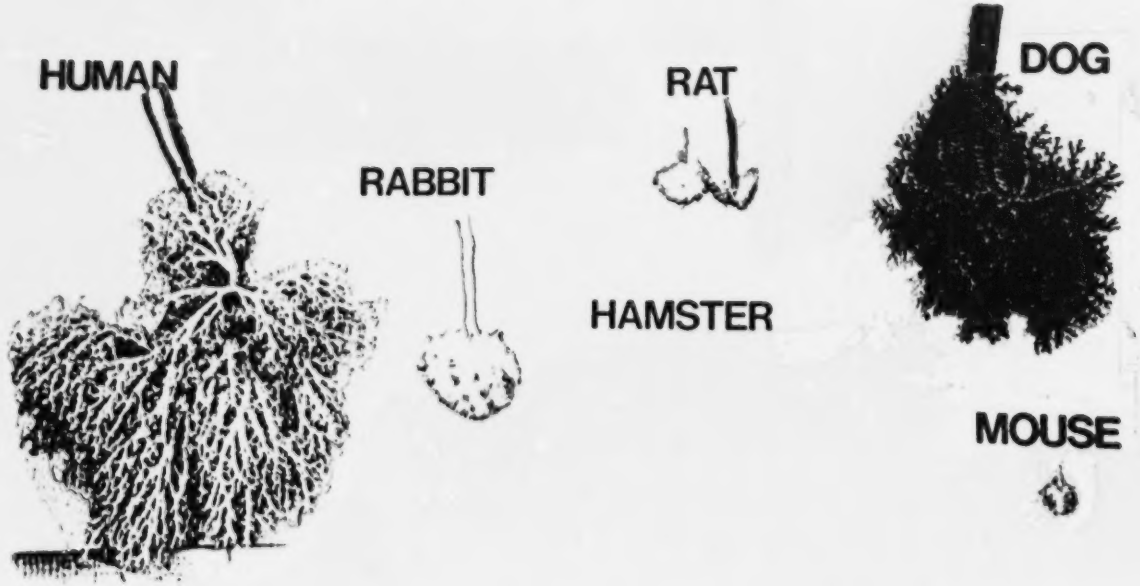

Figure 1. Silicone lung casts of different species.

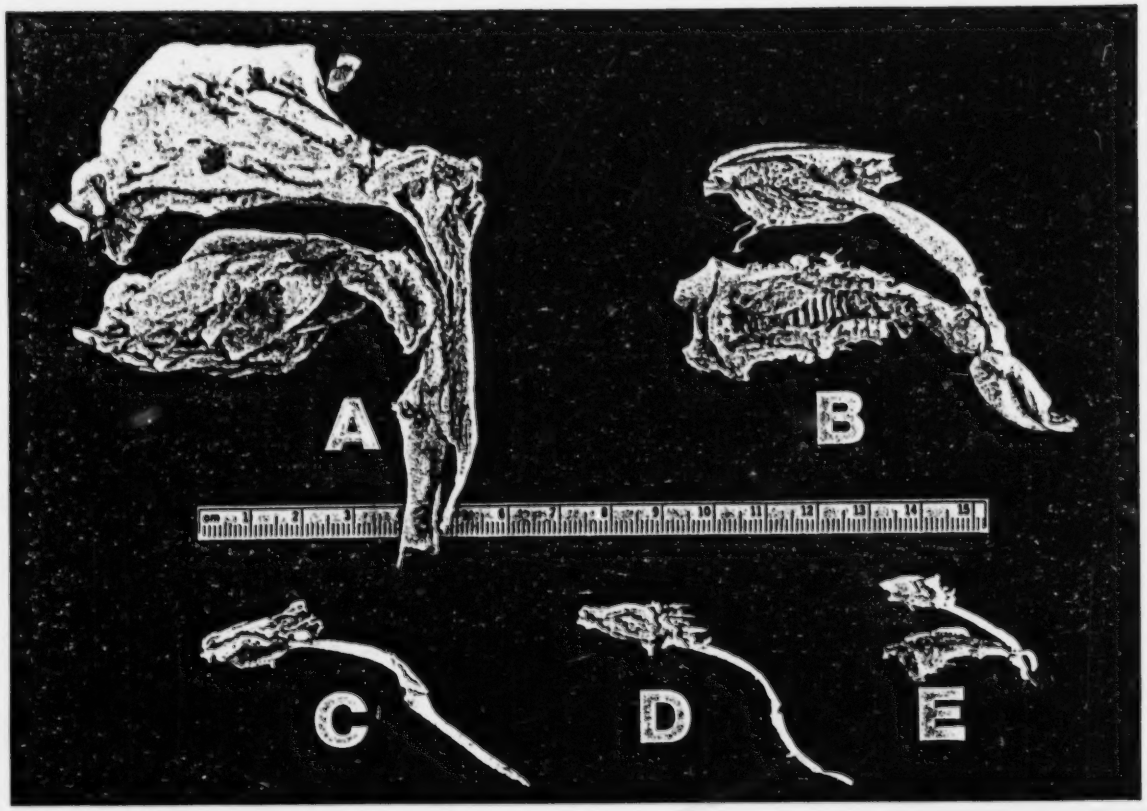

Figure 2. Silicone casts of the nasal-pharyngeal airway of different species: (a) human, (b) rabbit, (c) rat, (d) guinea pig, and (e) hamster.

TABLE 1. DESCRIPTIONS OF SPECIES USED IN THE STUDY

Species	Description	Remarks
Golden hamster	Male, adult, 150 g	Nasal measurements
Rat	Male, adult (C-D), 253 g	Nasal and lung airway measurements
Dog	Male, mongrel, 15 kg	Lung airway measurements
Baboon	Male, adult, 30 kg	Lung airway measurements
Human	Female, child (3 yr old), 13.6 kg	Nasal measurements
Human	Male, adult (39 yr old), 99.8 kg	Nasal and lung airway measurements
Rabbit	Male, adult, 2.2 kg	Nasal measurements
Guinea pig	Female, adult, 480 g	Nasal measurements

TABLE 2. COMPARATIVE LUNG MORPHOMETRIC DATA FROM DIFFERENT LUNG SPECIMENS

Lung Section	Lung Dimensions											
	Human, Adult[a] Age: 54 yr Body wt.: 81 kg Male			Baboon, Adult[b] Age: Unknown Body wt.: 30 kg Male			Mongrel Dog, Adult Age: Unknown Body wt.: 15 kg Male			C-D Rat, Adult[c] Age: 30 d Body wt.: 251 g Male		
	L (cm)	D (cm)	θ[d] (degree)	L (cm)	D (cm)	θ[d] (degree)	L (cm)	D (cm)	θ[d] (degree)	L (cm)	D (cm)	θ[d] (degree)
Trachea (zero generation)	12.0	2.01	0	16.83	1.60	0	21.6	2.00	0	2.84	0.25	NA
Main bronchus (first generation)	4.36	1.56	33	2.25	1.21	31	2.07	1.70	37	0.41	0.20	NA
Lobar bronchus (second generation)	1.78	1.13	34	1.39	0.99	24	1.49	1.16	20	0.35	0.16	NA
Segmental bronchus (third generation)	0.96	0.83	22	0.93	0.62	23	1.36	0.87	18	0.18	0.13	NA
Sub-segmental bronchus I (fourth generation)	0.99	0.65	20	0.71	0.31	32	0.90	0.62	43	0.12	0.09	NA
Sub-segmental bronchus II (fifth generation)	1.01	0.57	18	0.92	0.45	31	0.69	0.42	72	0.09	0.07	NA

NA = not available.
L = length of lung.
D = diameter of lung.
[a]From published data, Reference 15.
[b]From published data, Reference 31.
[c]From published data, Reference 27.
[d]The branching angle indicates the angular change in direction from the parent branch.